

# EFFECT OF MAGNESIUM ON BENEFICIAL ORGANISMS

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## Abstract

The paper presents the results of research on magnesium effect on beneficial entomopathogenic fungi and nematodes as well as some predatory arthropods.

Magnesium fertilization of soil contaminated with heavy metals does not significantly affect numbers of the majority of epigeal invertebrates, but it may influence numbers of single species, favouring the occurrence of some (*Bembidion* sp.) while restricting the presence of others (*Harpalus rufipes* De Geer).

Magnesium synergism with heavy metal ions has been found to increase infectiveness and pathogenicity of entomopathogenic fungi. Magnesium, in a dose of  $160 \text{ mg} \cdot \text{dm}^{-3}$  present in the medium, significantly enhances pathogenicity of fungi, such as *Beauveria bassiana*, *Paecilomyces farinosus*, *Paecilomyces fumoso-roseus* or *Metarhizium ansopliae*. Increased pathogenicity of *Steinernema carpocapsae* and *Heterorhabditis bacteriophora* towards test insects has also been observed when magnesium ions were added to a solution in which these nematodes were kept. An effective magnesium dose differed depending whether the nematodes were used separately for test insects ( $450 \text{ mg} \cdot \text{dm}^{-3}$ ) or jointly with entomopathogenic fungi ( $320 \text{ mg} \cdot \text{dm}^{-3}$ ). This protective effect of magnesium ions on beneficial microorganisms has also been observed in soil contaminated with heavy metals. An addition of magnesium to a solution in which entomopathogenic nematodes were kept ( $160 \text{ mg} \cdot \text{dm}^{-3}$ ) and to a medium on which fungi were cultured ( $320 \text{ mg} \cdot \text{dm}^{-3}$ ) increased pathogenic abilities of these organisms in contaminated soil to a very high degree (10- to 300- fold higher than the natural heavy metal content in soil).

**Keywords:** magnesium, epigeal fauna, entomopathogenic fungi, entomopathogenic nematodes.

## WPŁYW MAGNEZU NA WYBRANE ORGANIZMY POŻYTECZNE

## Abstrakt

W pracy zestawiono wyniki badań nad wpływem magnezu na pożyteczne grzyby i nicienie owadobójcze oraz wybrane stawonogi drapieżne.

Nawożenie magnezowe gleby skażonej metalami ciężkimi nie wpływa istotnie na liczebność większości grup bezkręgowców naziemnych, może jednak wpływać na liczebność pojedynczych gatunków, sprzyjając występowaniu jednych (*Bembidion* sp.), a ograniczając występowanie innych (*Harpalus rufipes* De Geer).

Stwierdzono synergizm magnezu z jonami metali ciężkich w podnoszeniu infekcyjności i patogeniczności grzybów owadobójczych. Magnez obecny w pożywce, w dawce  $160 \text{ g-dm}^{-3}$ , istotnie podwyższał patogeniczność takich grzybów, jak: *Beauveria bassiana*, *Paecilomyces farinosus*, *Paecilomyces fumoso-roseus*, *Metarhizium ansopliae*. Zaobserwowano również zwiększenie patogeniczności nicieni owadobójczych *Steinernema carpocapsae* i *Heterorhabditis bacteriophora* wobec owadów testowych po dodaniu jonów magnezu do roztworu, w którym przechowywano te nicienie. Efektywna dawka magnezu była zróżnicowana, zależnie od tego, czy nicienie stosowano samodzielnie wobec owadów testowych ( $450 \text{ mg-dm}^{-3}$ ), czy też wraz z grzybami owadobójczymi ( $320 \text{ mg-dm}^{-3}$ ).

Obserwowano także protekcyjny wpływ jonów magnezu na pożyteczne mikroorganizmy w warunkach gleby zanieczyszczonej metalami ciężkimi. Dodanie magnezu do roztworu, w którym przechowywano nicienie owadobójcze ( $160 \text{ mg-dm}^{-3}$ ), i do pożywki, na której hodowano grzyby ( $320 \text{ mg-dm}^{-3}$ ), zwiększało zdolności patogeniczne tych organizmów w warunkach gleby skażonej metalami ciężkimi nawet w bardzo wysokim stopniu (10-300-krotnie wyższa zawartość metali ciężkich w glebie ponad zawartość naturalną).

Słowa kluczowe: magnez, fauna naziemna, grzyby entomopatogeniczne, nicienie entomopatogeniczne.

## INTRODUCTION

Both representatives of beneficial epigeal entomofauna (*Carabidae*, *Staphylinidae*, *Arachnida*) and beneficial microorganisms (entomopathogenic fungi and nematodes, such as: *Beauveria bassiana*, *Paecilomyces farinosus*, *Paecilomyces fumoso-roseus*, *Metarhizium ansopliae*, *Steinernema carpocapsae* and *Heterorhabditis bacteriophora*) are an important factor allowing to maintain populations of many harmful insects at a level which does not pose any threat to agronomic crops. The efficiency of these organisms may be modified by the effect of soil anthropogenic pollution such as heavy metals (JAWORSKA et al. 1996, JAWORSKA et al. 1997 a, c). Frequently, besides liming, magnesium fertilization of soils polluted with heavy metals is mentioned as a means to diminish their toxicity. Magnesium may be used for simple chemical stimulation of the pathogenicity of beneficial microorganisms.

This paper presents a compilation of the results of research on the effect of magnesium fertilization of soil contaminated with heavy metals on beneficial epigeal fauna and investigations on potential practical enhancement of the effectiveness of entomopathogenic fungi and nematodes by means of their conditioning with magnesium.

## MATERIALS AND METHODS

The paper comprises results of field and laboratory analyses. The field experiment was conducted in 2000 on an arable field in Zagaje Stradowskie, a village in the Świętokrzyskie Province. Broad beans (*Vicia faba* L. ssp. *maior*), cv. White Windsor was cultivated on the following objects:

1. Control (soil with natural content of heavy metals: 8.2 mg Cu, 52.9 mg Zn, 28.2 mg Ni and 0.6 mg Cd·kg<sup>-1</sup>d.m. soil);
2. Soil contaminated with heavy metals dosed: 2 mg Cd, 15 mg Ni, 30 mg Cu, 70 mg Zn and 80 mg Pb·kg<sup>-1</sup> d.m. soil);
3. Soil receiving magnesium fertilizer (50 kg MgO·ha<sup>-1</sup>) and contaminated with heavy metals;
4. Soil receiving magnesium fertilizer (50 kg MgO·ha<sup>-1</sup>) and liming (2900 kg CaO·ha<sup>-1</sup>), contaminated with heavy metals.

Analyses on enthomopathogenic fungi and nematodes were conducted under laboratory conditions using various concentrations of magnesium in media for fungi culturing or in aqueous solutions for keeping nematodes. The methods were presented in detail in other papers (JAWORSKA, GOSPODAREK 2003 a, b, JAWORSKA et al. 1996, JAWORSKA et al. 1999).

## RESULTS AND DISCUSSION

Soil contamination with heavy metals causes a decrease in the abundance of captured epigeal fauna (Figure 1). Magnesium fertilization and liming of contaminated soil does not significantly influence the counts of most analyzed invertebrate groups, but it may affect the numbers of individual species. In the case of small *Bembidion* (Col., *Carabidae*) beetles a slight increase in the number of trapped insects may be observed in soil contaminated with heavy metals after application of magnesium fertilization. On the other hand, a common *Carabidae Harpalus rufipes* De Geer species, which occurred as numerous in soil contaminated with heavy metals as in the control, was considerably less often trapped on the magnesium fertilized object. The data on the effect of liming on epigeal fauna are quite divergent. In some research, the number of trapped epigeal fauna after liming of the polluted soil increased. Differences in the response to various degrees of farming intensity were also noted among various *Carabidae* species (JAWORSKA 1997, PAŁOSZ 1998). On fields under the most intensive farming system, fewer *Carabidae* were trapped. The species which most strongly decreased in number under more intensive insecticidetreatments included *Carabus auratus* L., *Pterostichus cupreus* L., *Pterostichus melanarius* III. and *Amara* sp. On the other hand, *Pterostichus lepidus* Leske species was more

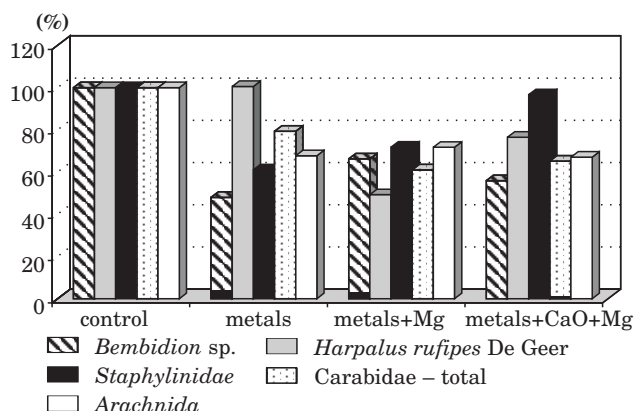


Fig. 1. Representatives of beneficial epigeal fauna in soil with natural heavy metal content and in metal contaminated soil receiving magnesium treatment (% in relation to the control)

numerously caught on a field where more intensive chemical protection was used. The latter fact was explained by the authors as a result of diversified phenology of this species occurrence and therefore its smaller exposure to pesticide poisoning. In the above research both *Ophonus rufipes* De Geer species and *Bembidion* sp. genus were significantly less numerous trapped on the most intensively farmed fields in comparison with extensive tillage, although a greater (3-fold) decline was found for *Bembidion* sp. beetles.

Entomopathogenic fungi are quite tolerant to heavy metal presence in the environment (ROUX et al.1993). The limits of this tolerance may change under the influence of interaction between metal ions (JAWORSKA, TOMASIK 1999). Investigations on the effect of various metals on the growth of entomopathogenic *Paecilomyces fumoso-roseus* fungus colonies and its infectiveness towards *Bruchus pisorum* L. evidenced a marked (ca 25%) increase in infectiveness of this fungus owing to 160 mg·dm<sup>-3</sup> supplement of Mg ions added to the medium (Table 1). The effect occurred despite the fact that magnesium did not reveal positive influence on the growth of fungal colonies. The result of combining Mg with other metals on the fungus was also investigated. Various metal ions previously added to the medium caused an inhibition of fungal colony growth and, in most cases, a decline in its infectiveness. On the other hand, a supplement of magnesium ions added to media which had already contained single metals (Al, Fe, V, Cr or Pb) alleviated the toxic effect and enhanced the infectiveness of fungi as compared with the control.

The research on potential chemical stimulation of entomopathogenic activity of other fungal species towards a legume pest *Sitona lineatus* L. demonstrated that Mg ion in the medium (160 mg·dm<sup>-3</sup>) significantly raised pathogenicity of such fungi as *Beauveria bassiana*, *Paecilomyces farinosus*, *Paecilomyces fumoso-roseus* or *Metarhizium ansopliae* (Table 1).

Table 1

Effect of mangesium on enthomopathogenic fungi and nematodes

Investigated feature	Control	Mg
Mortality (%) of <i>Bruchus pisorum</i> L. on the 5 <sup>th</sup> day after its contact with <i>P. fumosoroseus</i> grown on agar broth with added Mg ions (160 mg·dm <sup>-3</sup> )	73.30 b*	93.30 a
Mortality (%) of <i>Sitona lineatus</i> L. on the 5 <sup>th</sup> day after its contact with enthomopathogenic fungi grown on a media with Mg ions (160 m·dm <sup>-3</sup> ): <i>Beauveria bassiana</i> <i>Paecilomyces farinosus</i> <i>Paecilomyces fumoso-roseus</i> <i>Metarhizium ansopliae</i>	46.60 a 36.60 a 36.60 a 43.30 a	100.0 b 66.70 b 50.00 a 63.30 b
Mortality (%) of <i>Galleria mellonella</i> on the 5 <sup>th</sup> day after its contact with enthomopathogenic fungi and nematodes grown on a medium with Mg ions (mg·dm <sup>-3</sup> ): <i>Beauveria bassiana</i> Mg 450 Mg 320 Mg 160 <i>Steinernema carpocapsae</i> Mg 450 Mg 320 Mg 160 <i>S. carpocapsae</i> + <i>B. bassiana</i> Mg 450 Mg 320 Mg 160	24.20 a    65.10 a   70.20 ab	    38.70 a 71.30 a 62.10 a  95.10 b 76.4 0 ab 81.00 ab  87.60 ab 99.00 b 78.80 a
Effect of conditioning nematodes in aqueous solutions of Mg (160 mg·dm <sup>-3</sup> ) ions on their pathogenicity (mortality (%)) of <i>Sitona lineatus</i> L. on the 7th day after its contact with enthomopathogenic nematodes) <i>Steinernema carpocapsae</i> <i>Heterorhabditis bacteriophora</i>	75.10 a 47.40 a	100.0 b 91.70 b
Effect of conditioning nematodes in aqueous solutions of Mg (160 mg·dm <sup>-3</sup> ) ions on their reproduction inside the <i>S. lineatus</i> body (number of nematode invasive larvae per mg body weigth) <i>Steinernema carpocapsae</i> <i>Heterorhabditis bacteriophora</i>	77.00 a 111.0 a	144.0 b 184.0 b
Mortality (%) of <i>Galleria mellonella</i> on the 2 <sup>nd</sup> day after its contact with enthomopathogenic nematodes (conditioned in aqueous solutions of Mg (mg·dm <sup>-3</sup> ): <i>Steinernema carpocapsae</i> 32 320 <i>Heterorhabditis bacteriophora</i> 32 320	80 a   80 a	100 b 100 b  100 b 100 b

\*Means for each property marked with the same letter do not differ at  $P > 0.05$ .

Heavy metals, such as cadmium, copper, zinc and lead, cause increased mortality of enthomopathogenic nematode infective juveniles as well as decrease their pathogenicity towards test insects and reproduction (JAWORSKA, GORCZYCA 2002). In an experiment using soil strongly contaminated with heavy metals, decreased infectiveness of these beneficial microorganisms was found (ROPEK, GORCZYCA 2000). Analogously to enthomopathogenic fungi, a beneficial effect of magnesium ions was found for nematodes, visible as their increased pathogenicity (JAWORSKA et al. 1996, JAWORSKA et al. 1997 a,b). In experiments on stimulating enthomopathogenic activity of fungi and nematodes applied jointly on *Galeria mellonella* test insects it was found that, among three tested Mg ion concentrations, the concentration of  $320 \text{ mg} \cdot \text{dm}^{-3}$  increased most infectiveness of the analyzed microorganisms. When only *Steinernema carpocapsae* nematodes were used for the test insects, the highest mortality of *Galeria mellonella* caterpillars was obtained at the highest Mg ion concentration ( $450 \text{ mg} \cdot \text{dm}^{-3}$ ) used for the nematode stimulation (Table 1). An advantageous effect of keeping *S. carpocapsae* and *Heterorhabditis bacteriophora* nematode in aqueous Mg solution ( $160 \text{ mg} \cdot \text{dm}^{-3}$ ) was also found when various development stages of *Sitona lineatus* L. were controlled using these organisms (Table 1). In this case, magnesium acted positively by increasing reproduction of the nematode species.

The experiments on magnesium interaction with heavy metal ions added to soil towards enthomopathogenic fungi and nematodes revealed that magnesium supplement to a solution used for keeping nematodes ( $160 \text{ mg} \cdot \text{dm}^{-3}$ ) and to a medium for fungi ( $320 \text{ mg} \cdot \text{dm}^{-3}$ ) enhanced pathogenicity of both organisms in soils with heavy metals 10- to 300-fold exceeding their natural content (JAWORSKA 1999).

## CONCLUSIONS

1. Magnesium fertilization of soil contaminated with heavy metals does not affect significantly the number of most epigeal invertebrate groups, although it may influence single species, favouring some (*Bembidion* sp.) but limiting the occurrence of others (*Harpalus rufipes* De Geer).

2. A supplement of magnesium ions added to a solution used for keeping enthomopathogenic nematodes or to a medium for fungal cultures may be recommended as a simple method to stimulate biological activity of these microorganisms.

3. Magnesium can also protect the above microorganisms in soil polluted with heavy metals.

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